

U.S.S.N. 08/126,505

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SUPPLEMENTAL AMENDMENT

Claims Listing

1. (Currently Amended) An analog of a protein-A modified form of a regulator of complement activation protein (RCA protein) wherein the RCA protein is selected from the group consisting of complement receptor 1, complement receptor 2, decay accelerating factor, membrane cofactor protein, C4 binding protein, factor H, and these these complement regulating RCA proteins wherein the carboxy terminus of the RCA protein is removed to allow the protein to be secreted, wherein said the modified form protein analog is selected from the group consisting of:

- a) a hybrid RCA protein complement regulating protein analogs containing comprising short consensus repeats (SCRs) derived from a second two, different complement regulating protein of the RCA proteins not including combinations consisting of complement receptor 1 and complement receptor 2,
- b) a recombined RCA protein complement regulating protein analogs wherein the short consensus repeats-SCRs of the RCA protein are rearranged, and
- c) a truncated RCA protein complement regulating protein analogs consisting of as few as three SCRs short consensus repeats,

wherein the modified form of the RCA protein analog binds C3b, C4b or C3b and C4b.

2. (cancelled)

2. ~~2.~~ (Currently Amended) The analog modified form of the RCA protein of claim 1 wherein the RCA protein is complement receptor one 1.

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3 ~~4~~. (Currently Amended) The ~~analog~~ modified form of an RCA protein of claim 1 wherein the RCA protein is decay accelerating factor.

4 ~~5~~. (Currently Amended) The ~~analog~~ modified form of the RCA protein of claim 1 wherein the RCA protein is factor H.

6. (cancelled)

7. (cancelled)

5 ~~8~~. (Currently Amended) ~~An analog of a protein-A~~ modified form of an RCA protein wherein the RCA protein is selected from the group consisting of complement receptor 1, complement receptor 2, decay accelerating factor, membrane cofactor protein, C4 binding protein, factor H, and these RCA proteins wherein the carboxy terminus of the RCA protein is removed to allow the protein to be secreted, wherein the ~~protein-analog~~ modified form contains amino acid substitutions in the ~~short consensus repeats~~ SCRs which correspond to amino acid substitutions in the ~~short consensus repeats~~ SCRs of complement receptor one 1 (SEQ ID No: 13) selected from the group consisting of:

CR1-4 with its first 122 amino acids (SCR1-2) (Sequence ID Nos 1 and 3) replaced with CR1 amino acids 497-618 (SCR 8-9) (Sequence ID Nos. 2 and 4) and CR1-4(8,9) with deletion of 194-253; and substitution of amino acids 271-543 with: T-R-T-T-F-H-L-G-R-K-C-S-T-A-V-S-P-A-T-T-S-E-G-L-R-L-C-A-A-H-P-R-E-T-G-A-L-Q-P-P-H-V-K (Sequence ID No. 11), or these amino acid sequences where any I is replaced with either L or V, any L is replaced with either I or V, any V is replaced with I, L, or F, any F is replaced with V, any K is replaced with

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R, any R is replaced with K, any Q is replaced with N, any N is replaced with Q, any D is replaced with E[[.]], any E is replaced with D, any G is replaced with A, or any A is replaced with G.

9. (cancelled)

~~10.~~ 10. (Currently Amended) ~~An analog~~ A modified form of decay accelerating factor wherein one or more substitutions are introduced into the region of the protein corresponding to decay accelerating factor ~~short consensus repeats~~ SCRs 2-3 as shown in Sequence ID No. 17 selected from the group consisting of 180-187: S-T-K-P-P-I-C-Q (amino acids 54-61 of Sequence ID No. 4); 175-178: N-A-A-H (amino acids 49-52 of Sequence ID No. 4); 175-187: S-T-K-P-P-I-C-Q-N-A-A-H (Sequence ID No. 9); 130: R (amino acid 4 of Sequence ID No. 3); 145: D (amino acid 19 of Sequence ID No. 4); 77-84: K-L-K-T-Q-T-N-A-S-D (amino acids 12-21 of Sequence ID No. 2); 90-92: S-L-K (amino acids 27-29 of Sequence ID No. 2), and these amino acid sequences where I is replaced with either L or V, L is replaced with either I or V, V is replaced with I, L, or F, F is replaced with V, K is replaced with R, R is replaced with K, Q is replaced with N, N is replaced with Q, D is replaced with E, E is replaced with D, G is replaced with A, or A is replaced with G.

~~11.~~ 11. (Currently Amended) The ~~analog~~ modified form of the RCA protein of claim 1 wherein the ~~complement-regulatory~~ modified form of the RCA protein is factor H comprising sequences conferring on the modified form of the RCA protein an activity selected from the group consisting of Cab binding activity, C3b cofactor activity, C4b binding activity, and C4b

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cofactor activity, wherein the sequences are derived from a protein selected from the group consisting of complement receptor 1, membrane cofactor protein, C4 binding protein, and factor H.

8 ~~12~~. (Currently Amended) The ~~analog~~ modified form of the RCA protein of claim 1 ~~comprising wherein the hybrid RCA protein at least one comprises short consensus repeat~~ SCRs derived from ~~a different an RCA protein~~ selected from the group consisting of complement receptor 1, ~~complement receptor 2~~, decay accelerating factor, membrane cofactor protein, C4 binding protein, and factor H.

9 ~~13~~. (Currently Amended) The ~~analog~~ modified form of the RCA protein of claim 1 wherein the modified form of the RCA protein ~~analog~~ includes SCRs 2, 3 and 4 of DAF and has C3b cofactor activity, C4b cofactor activity and decay accelerating activity.

10 ~~14~~. (Currently Amended) The ~~analog~~ A modified form of the RCA protein of claim 1 wherein the truncated RCA protein ~~region of the protein having biological activity~~ consists of three ~~short consensus regions~~ SCRs and has two complement regulatory activities.

11 ~~15~~. (Currently Amended) The ~~analog of claim~~ modified form of the RCA protein of claim 1, ~~3~~ ~~8~~ 4 or 12, further comprising a pharmaceutically acceptable carrier ~~for administration to a patient in need thereof.~~

12 ~~16~~. (Currently Amended) A method for making ~~an analog of a protein~~ a modified form of an RCA protein wherein the RCA protein is selected from the group consisting of complement receptor 1, ~~complement receptor 2~~, decay accelerating factor, membrane cofactor protein, C4

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binding protein, factor H, and these ~~complement-regulating~~ RCA proteins wherein the carboxy terminus of the RCA protein is removed to allow the protein to be secreted, ~~comprising~~ constructing a DNA sequence encoding a protein analog wherein the modified form is selected from the group consisting of:

- a) a hybrid RCA protein ~~complement-regulating protein analogs~~ containing comprising short consensus repeats SCRs derived from a second two, different complement-regulating protein of the RCA proteins not including combinations consisting of complement receptor 1 and complement receptor 2,
- b) a recombined RCA protein ~~complement-regulating protein analogs~~ wherein the short consensus repeats SCRs of the RCA protein are rearranged, and
- c) a truncated RCA protein ~~complement-regulating protein analogs~~ consisting of as few as three SCRs short consensus repeats,

wherein the modified form of the RCA protein analog binds C3b, C4b, or C3b and C4b, and

the method comprising expressing the a DNA sequence encoding the modified form of the RCA protein in a suitable host cell for expression of the protein analog.

17. (cancelled)

13 ¹² 18. (Currently Amended) The method of claim ~~16~~ wherein the RCA protein ~~used to~~ form the analog is complement receptor ~~one~~ 1.

14 ¹² 19. (Currently Amended) The method of claim ~~16~~ wherein the RCA protein is decay accelerating factor.

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15 20. (Currently Amended) The method of claim ¹²16 wherein the RCA protein is factor H.

21. (cancelled)

22. (cancelled)

16 23. (Currently Amended) A method for making an analog of a protein ~~a modified form~~ of an RCA protein wherein the RCA protein is selected from the group consisting of complement receptor 1, complement receptor 2, decay accelerating factor, membrane cofactor protein, C4 binding protein, factor H, and these proteins wherein the carboxy terminus of the RCA protein is removed to allow the protein to be secreted, wherein the protein analog modified form of an RCA protein contains amino acid substitutions in the ~~short consensus repeats~~ SCRs which correspond to amino acid substitutions in the ~~short consensus repeats~~ SCRs of complement receptor ~~one~~ 1 (SEQ ID No: 13) selected from the group consisting of:

CR1-4 with its first 122 amino acids (SCR1-2) (Sequence ID Nos. 1 and 3) replaced with CR1 amino acids 497-618 (SCR 8-9) (Sequence ID Nos. 2 and 4) and CR1-4(8,9) with deletion of 194-253; substitution of amino acids 271-543 with: T-R-T-T-F-H-L-G-R-K-C-S-T-A-V-S-P-A-T-T-S-E-G-L-R-L-C-A-A-H-P-R-E-T-G-A-L-Q-P-P-H-V-K (Sequence ID No. 11), and these amino acid sequences where any I is replaced with either L or V, any L is replaced with either I or V, any V is replaced with I, L, or F, any F is replaced with V, any K is replaced with R, any R is replaced with K, any Q is replaced with N, any N is replaced with Q, any D is replaced with E, any E is replaced with D, any G is replaced with A, or any A is replaced with G,

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the method comprising expressing a DNA encoding the ~~protein analog~~ modified form of the RCA protein in a ~~suitable~~ host cell ~~and recovering the protein analog~~.

17 24. (Currently Amended) A method for making ~~an analog of a protein~~ a modified form of an RCA protein wherein the RCA protein is selected from the group consisting of complement receptor 1, complement receptor 2, decay accelerating factor, membrane cofactor protein, C4 binding protein, factor H, and these proteins wherein the carboxy terminus of the RCA protein is removed to allow the protein to be secreted, wherein the ~~protein analog~~ modified form of an RCA protein contains amino acid substitutions in the ~~short consensus repeats~~ SCRs which correspond to amino acid substitutions in the ~~short consensus repeats~~ SCRs of complement receptor ~~one~~ 1 (SEQ ID No: 13) selected from the group consisting of:

79: D (amino acid 19 of Sequence ID No. 4); 37,79: Y,D (amino acid 37 of Sequence ID No. 2 and amino acid 19 of Sequence ID No. 4); 92: T (amino acid 32 of Sequence ID No. 4); 92-94: K...Y (amino acids 32-34 of Sequence ID NO. 3); 99,103,106: S...T...I (amino acids 39, 43 and 46 of Sequence ID No. 3); 109-112: D-T-V-I (amino acids 49-52 of Sequence ID No. 3); 110: T (amino acid 50 of Sequence ID No. 3); 111: V (amino acid 51 of Sequence ID No. 3); 112: I (amino acid 52 of Sequence ID No. 3); 1,3: Q...N (amino acids 1, 3 of Sequence ID No. 1); 6-9: E-W-L-P (amino acids 6-9 of Sequence ID No. 1); 12-16, 18-21: K-L-K-T-Q...N-A-S-D (amino acids 12-21 of Sequence ID No. 2); 27,29: S...K (amino acids 27,29 of Sequence ID No. 2); 37: S (amino acid 37 of Sequence ID No. 1); 44, 47, 49: I...K...S (amino acids 44, 47, 49 of Sequence ID No. 1); 52-54, 57, 59: TG-A...R...R (amino acids 52-54, 57, 59 of Sequence ID No.

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1); 78-79, 82: K-G ...F (amino acids 18-19, 22 of Sequence ID No. 3); 85, 87: Q...K (amino acids 25, 27 of Sequence ID No. 3); 12-16, 18-21: R-P-T-N-L ...D-E-F-E (amino acids 12-21 of Sequence ID No. 1); 27,29: Y...N (amino acids 27, 29 of Sequence ID No. 1); 35, 64-65, 94: G...R-N...Y (amino acid 35 of Sequence ID No. 1, amino acids 4-5, 34 of Sequence ID No. 3), and these amino acid sequences where any I is replaced with either L or V, any L is replaced with either I or V, any V is replaced with I, L, or F, any F is replaced with V, any K is replaced with R, any R is replaced with K, any Q is replaced with N, any N is replaced with Q, any D is replaced with E, any E is replaced with D, any G is replaced with A, or any A is replaced with G, the method comprising expressing a DNA encoding the protein analog modified form of the RCA protein in a suitable host cell and recovering the protein analog.

18 25. (Currently Amended) A method for making an analog a modified form of decay accelerating factor wherein one or more substitutions are introduced into the region of the protein corresponding to decay accelerating factor ~~short consensus repeats~~ SCRs 2-3 as shown in Sequence ID No. 17 selected from the group consisting of

180-187: S-T-K-P-P-I-C-Q (amino acids 54-61 of Sequence ID No. 4); 175-178: N-A-A-H (amino acids 49-52 of Sequence ID No. 4); 175-187: S-T-K-P-P-I-C-Q-N-A-A-H (Sequence ID No. 9); 130: R (amino acid 4 of Sequence ID No. 3); 145: D (amino acid 19 of Sequence ID No. 4); 77-84: K-L-K-T-Q-T-N-A-S-D (amino acids 12-21 of Sequence ID No. 2); 90-92: S-L-K (amino acids 27-29 of Sequence ID No. 2), and these amino acid sequences where any I is replaced with either L or V, any L is replaced with either I or V, any V is replaced with I, L, or F,

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any F is replaced with V, any K is replaced with R, any R is replaced with K, any Q is replaced with N, any N is replaced with Q, any D is replaced with E, any E is replaced with D, any G is replaced with A, or any A is replaced with G.

19 26. (Currently Amended) The method of claim ¹²16 wherein the ~~complement regulatory~~ RCA protein is factor H comprising sequences conferring on the protein an activity selected from the group consisting of C3b binding activity, C3b cofactor activity, C4b binding activity, and C4b cofactor activity, wherein the sequences are derived from ~~[[a]]~~ an RCA protein selected from the group consisting of complement receptor 1, membrane cofactor protein, C4 binding protein, and factor H.

20 27. (Currently Amended) The method of claim ¹²16 ~~comprising expressing a wherein~~ the DNA sequence encodes encoding a protein a hybrid RCA protein comprising SCRs derived from an RCA protein selected from the group consisting of complement receptor 1, complement receptor 2, decay accelerating factor, membrane cofactor protein, C4 binding protein and factor H, including in reading frame a DNA encoding at least one SCR-short consensus repeat derived from a different RCA protein selected from the group consisting of complement receptor 1, complement receptor 2, decay accelerating factor, membrane cofactor protein, C4 binding protein, and factor H, not including combinations consisting of complement receptor 1 and complement receptor 2.

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21 ¹² 28. (Currently Amended) The method of claim ¹² 16 wherein the ~~protein analog~~
modified form of the RCA protein includes SCRs 2, 3 and 4 of DAF and has C3b cofactor
activity, C4b cofactor activity and decay accelerating activity.

22 ¹² 29. (Currently Amended) The method of claim ¹² 16 wherein the ~~protein-modified form~~
of the RCA protein is a truncated RCA protein consisting of three SCRs ~~consists essentially of~~
~~three short consensus regions~~ and ~~has~~ having two complement regulatory activities.

23 ¹² 30. (Currently Amended) The method of claim ¹² 16 further comprising ~~isolated~~
isolating the analog modified form of the RCA protein and mixing with the isolated ~~analog~~
modified form of an RCA protein a pharmaceutically acceptable carrier ~~for administration to a~~
~~patient in need thereof.~~

24 ³ 31. (Currently Amended) A DNA sequence which encodes ~~an analog of claim a~~
modified form of the RCA protein of claim 1, ³ 4, or ⁸ 12.

25 ²⁴ 32. (Currently Amended) The DNA sequence of claim ²⁴ 31 inserted into an expression
vector operably linked to control sequences compatible with a host cell, which expression vector
is capable, when transformed into the host cell, of expressing a DNA encoding the ~~analog of~~
claim 1 modified form of the RCA protein.

33. (cancelled)

26 ³⁴ 34. (Currently Amended) A method for enhancing the C4b or C3b cofactor activity of
~~a complement regulatory~~ an RCA protein, wherein the protein has either C3b or C4b cofactor
activity, comprising adding sequences to the protein conferring binding of the other ligand, either

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C4b or C3b, wherein the sequences are present in a protein selected from the group of naturally occurring complement receptor 1, ~~complement receptor 2~~, decay accelerating factor, membrane cofactor protein, C4 binding protein, and factor H, ~~not including combinations consisting of complement receptor 1 and complement receptor 2.~~